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TECHNETIUM-99m MINICOLLOID
FOR RADIONUCLIDE LYMPHOGRAPHY

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Research was conducted according to the principles enunciated in the "Guide for Laboratory Animal Facilities and Care," prepared by the National Academy of Sciences - National Research Council.

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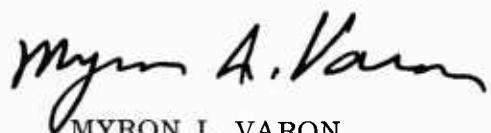
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FOREWORD
(Nontechnical summary)

Conventional radiographic lymphography has been shown to play a significant role in the initial diagnosis, treatment planning, and follow-up of many diseases. However, it remains a relatively difficult, time-consuming technique. It is also associated with definite morbidity and numerous potential complications. These factors have all motivated the search for an alternative diagnostic method for the study of the lymphatic system.

One approach has been the investigation of radiopharmaceuticals for lymphatic system imaging. Gold-198 radiocolloid has been used but has not found general acceptance. With the development of technetium-99m, it seemed evident that this isotope should be evaluated for use in radionuclide lymphography.

The objective of this study was to determine the relative efficacy of a recently developed radiopharmaceutical, technetium-99m "minicolloid", compared to a commercially available technetium-99m colloid for imaging the lymphatic system. Other purposes of this study were to determine the optimum dose to scan time for the radiopharmaceuticals, the effects of exercise on migration of the radiopharmaceutical through the lymphatic system, and the effects of the use of multiple injection sites on uptake of the radiopharmaceutical into the lymphatic channels. The use of local anesthetics and hyaluronidase was also considered.

Technetium-99m sulfur colloid was prepared utilizing the hydrogen sulfide method. Technetium-99m minicolloid is currently used as an effective bone marrow scanning agent. The physical characteristics of the technetium-99m isotope

were found to be more ideal for effective lymphatic imaging than gold-198. This new colloid material, referred to as minicolloid because of its relatively small particle size, gave much better lymph node and lymph channel visualization than the commercially available colloid. The study also revealed better radiopharmaceutical uptake in the nodal system, which enabled the study of the lymphatic system on the same day of radiopharmaceutical administration. Dose to scan time was therefore short, thus allowing these nuclear medicine procedures during a single patient visit.

ABSTRACT

The technique for lymphatic system evaluation by imaging with technetium-99m "minicolloid" is presented. Results from studies which compared the smaller particle radiocolloid with the commercially available radiopharmaceutical of the larger colloid particle size demonstrated the former to be a far superior agent for imaging the lymphatic system. This appears to be due to its more favorable migration through successive nodal groups. Other proven advantages of the minicolloid material were same-day imaging, lower absorbed dose, and better resolution. Radionuclide lymphography is expected to play an increasing role, in conjunction with radiographic lymphography, as larger numbers of patients suspected of lymphatic system disease are evaluated using the nuclear medicine techniques and approaches described.

I. INTRODUCTION

Conventional radiographic lymphography plays an important role in the initial diagnosis, treatment planning, and follow-up of many diseases.^{8,12} However, it remains a relatively difficult, time-consuming technique of limited repeatability. The procedure is associated with definite morbidity and numerous potential complications.⁷ It is contraindicated in patients with diminished cardiorespiratory reserve and those with a history of sensitivity to vital dyes, local anesthetics and iodinated contrast agents. These factors have all motivated the search for alternative diagnostic methods.

One approach has been the investigation of various radiopharmaceuticals for lymphatic system imaging. Initially gold-198 radiocolloid was used for radionuclide lymphography,^{6,11,13} but this agent has not found general acceptance. The ideal physical properties of technetium-99m, as described by Hauser et al.,⁵ suggested that ^{99m}Tc labeled colloid materials might prove to be ideal for radionuclide lymphography. Recently, Fairbanks et al.³ described the successful use of a commercially available sulfur colloid (Tesuloid from Squibb) for lymphatic imaging in a series of 31 patients. This suggested that ^{99m}Tc labeled sulfur colloid materials prepared by the H₂S method might prove to be ideal agents for lymphography.

While developing a clinically useful gamma scanning technique for our institution, the efficacy of the hydrogen sulfide preparation for lymphography (^{99m}Tc "mini-colloid" currently employed for bone marrow scanning in our institutions^{2,10}) was compared with that of certain commercial products. Other objectives of this study were to determine optimum dose to scan time, effects of exercise on migration of the

radiopharmaceuticals and effects of multiple injection sites. In addition, the use of local anesthetics and hyaluronidase was considered.

II. MATERIALS AND METHODS

The basic difference between ^{99m}Tc sulfur colloid prepared by the hydrogen sulfide method and the colloids now available commercially is particle size. This hydrogen sulfide preparation is called "minicolloid" to make this distinction. Particle size, as determined by light scatter spectrophotometry, is heterogeneous (less than 0.1 micron) and shows no discrete particle size for the minicolloid. The majority of the minicolloid particles passed readily through a 0.22-micron filter. A commercially available kit from New England Nuclear (NEN) yielded a particle size of 0.5 micron as 90 percent of the NEN colloidal particles did not pass through a 0.45-micron filter. Fairbanks et al.³ cited a particle size of 0.3 micron for the Tesuloid kits. Table I presents the stepwise procedure and key factors for successfully compounding ^{99m}Tc minicolloid.

The minicolloid and the NEN sulfur colloid were injected subcutaneously in the medial web spaces of the hind feet of six anesthetized beagle dogs. Approximately 1.5 mCi of each product were injected bilaterally. Serial counts were then obtained using a Nuclear-Chicago Pho/Gamma II HP camera. The disappearance of the radiopharmaceutical from the lower extremity was monitored by counting over the injection site and the popliteal lymph nodes. The appearance of the radiopharmaceutical in the pelvis and abdomen was monitored by counting over the iliolumbar nodal areas.

To study the effect of movement of the lower extremities on rate of radiopharmaceutical migration, relative counts were compared for two groups of animals, one group having its lower extremities exercised passively and the other being kept immobile.

Table I. Steps in Compounding Technetium-99m Minicolloid

<ol style="list-style-type: none">1. 15 - 20-ml beaker with stirring bar2. 2 - 8 ml ^{99m}Tc eluate3. 1-1/2 ml gelatin solution (10 mg/ml)4. 1 ml 12 N HCl (conc)5. Bubble in H_2S gas 5 min, stand 10 min6. Bubble in N_2 gas 5 min7. Check with lead acetate paper for absence of H_2S8. Adjust pH to 5.0 - 6.0 with 5 N and 1 N NaOH9. Filter through 0.22-micron membrane filter into sterile vial.

Key factors in successful compounding are:

<ol style="list-style-type: none">1. Make gelatin solution by placing 200 mg gelatin powder in a beaker, add 20 ml water for injection, heat to gentle boil for 3 min, cool, adjust volume to 20 ml, filter through 0.22-micron membrane filter into a series of sterile vials.2. Follow order of additions.3. Leave in acid state no longer than necessary.4. Carefully avoid letting pH rise above 6.0.

Finally, a preliminary evaluation in several human subjects was undertaken using a slightly modified comparative serial counting technique. Equivalent doses of 2.0 mCi of the minicolloid and the NEN colloid were injected in the medial web spaces of each foot. The pelvic and abdominal nodal areas (ilioinguinal and periaortic) were then serially counted separately. The injection sites were counted serially without including node-bearing areas. Subjects were asked to ambulate between counts. Additionally, four humans received injections in more than one web space on each side and some doses contained a local anesthetic.

III. RESULTS AND DISCUSSION

Comparison of minicolloid and NEN colloid. In the dog experiments, the minicolloid demonstrated more rapid hind extremity disappearance and iliolumbar appearance than did the NEN colloid (Figure 1). The shape of the iliolumbar appearance

curves is of interest. For both products, there was a rapid increase in count rate followed by a plateau during which little change in activity occurred. The plateau represents the optimum imaging period; and, significantly, the minicolloid activity was several times that of the NEN colloid. The later gradual increase in count rate reflects lymphatic vascular recirculation with radioactivity detected from the abdominal viscera.

Figure 2 is a scintigraphic expression of the importance of this observation. Figure 2a shows that 2 hours after bilateral injection of NEN colloid there is no activity beyond the popliteal nodes. Figure 2b, obtained 30 minutes after injection of

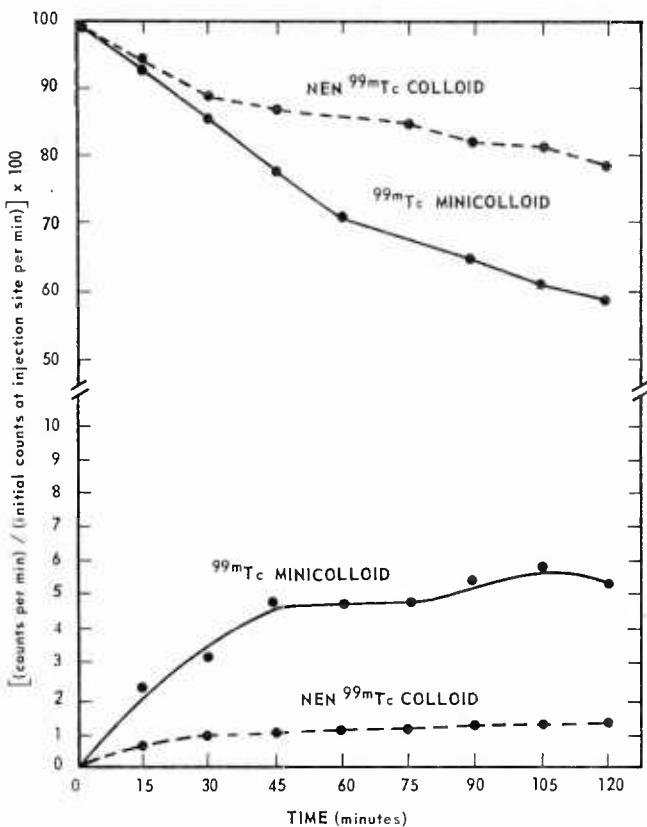


Figure 1. Kinetics for ^{99m}Tc minicolloid and NEN ^{99m}Tc colloid. The upper portion of the figure represents hind extremity disappearance curves. The lower portion of the figure represents iliolumbar appearance curves.

minicolloid on the left side of the same dog, shows clearly that lymphatic pathways and nodes on the left can be seen as high as the upper lumbar area.

The comparative results for minicolloid and NEN colloid in humans are summarized in Figure 3. Both radiopharmaceuticals disappeared equally promptly from the injection site. In humans, the first node encountered after injection into the medial web space is in the ilioinguinal region, and a difference of less than a factor of two existed between the relative uptake of the two products in these nodes. However, a manyfold difference was evident in the later uptake in the periaortic region. These

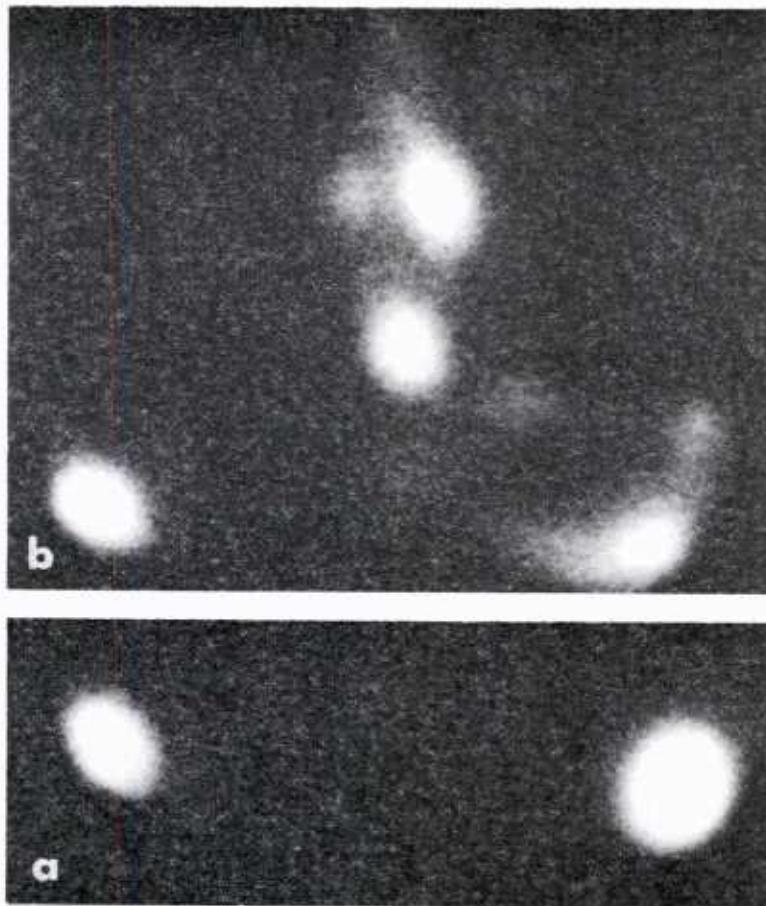


Figure 2. Scintiphotography of dog lymph nodes: (a) popliteal nodes after bilateral injection of NEN ^{99m}Tc colloid; (b) iliolumbar and periaortic nodes after subsequent unilateral injection of ^{99m}Tc minicolloid

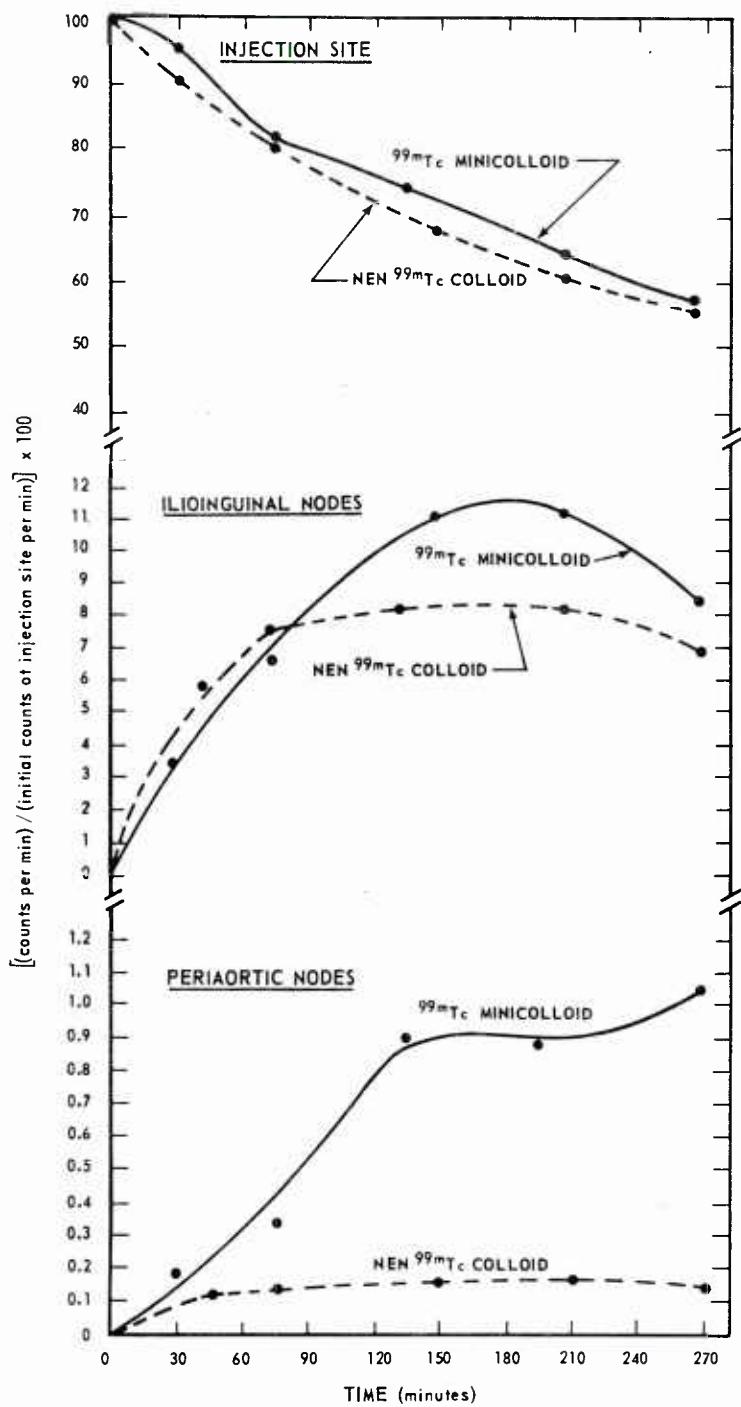


Figure 3. Results for minicolloid and NEN colloid in humans showing injection site disappearance curves and ilioinguinal and periaortic appearance curves

observations, coupled with the results from studies in dogs, indicate that the more favorable activity achieved with ^{99m}Tc minicolloid lies not in the more rapid initial uptake into the lymphatics but in the greater ability of the minicolloid to migrate through successive node levels. This is presumably due to its smaller particle size. The less favorable activity levels achieved with the larger particle colloid in the peri-aortic region are in agreement with Fairbanks et al.³ who stated that "more commonly the nodes above the external iliac group showed very faint uptake of ^{99m}Tc -colloid."

Exercise. The importance of exercise on the kinetics of ^{99m}Tc minicolloid in lymphatics is indicated by the results from dogs shown in Figure 4. In contrast to the rapid rise of iliolumbar activity seen with exercise, there was minimal activity evident without exercise, even by 3 hours after injection. Total iliolumbar activity remained several times less throughout the period of observation in the unexercised dogs.

Adjunctive factors. It was concluded that local anesthesia is not required for pedal injections. The discomfort after injection lasted no more than 10-15 seconds, a time shorter than onset of anesthetic effects. (Discomfort may persist for several minutes after injection in the hand and local anesthesia is recommended.) Hyaluronidase was not used and it was decided that it is not necessary with ^{99m}Tc minicolloid since uptake is rapid without it. As shown in this study, migrational limitations are not at the point of entry into the lymphatics but in the passage of colloid through successive node groups. By using a radiopharmaceutical with high specific activity, without hyaluronidase or a local anesthetic, the injection volume is small (0.2-0.5 ml per injection) and may be better tolerated by patients. Dose preparation is simpler when fewer drugs are involved.

It has been shown in radiographic studies that not all of the retroperitoneal nodes are visualized from any single injection.¹ The usual route of injection is into the greater saphenous system. Two or more injections into the distribution of this system with either the radiographic or the radioisotopic technique have no advantage over a single injection. Although a slight theoretical advantage would result from a second injection in the lesser saphenous system, we have not chosen to use this approach. Our technique involves the use of a single injection site in the medial web space of each foot.

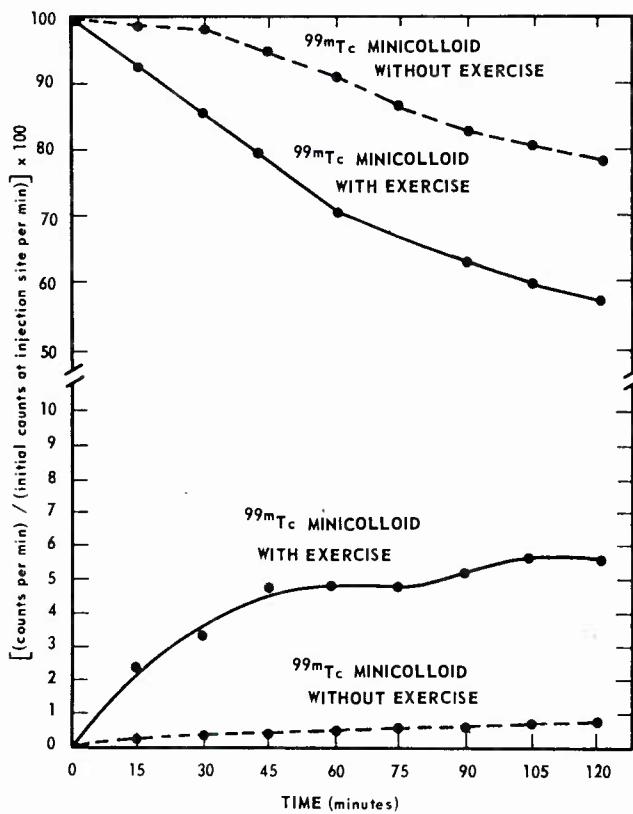


Figure 4. Effects of exercise with the ^{99m}Tc minicolloid. The upper portion of the figure represents hind extremity disappearance curves. The lower portion of the figure represents iliolumbar appearance curves.

The scintiphotos were obtained with a Nuclear-Chicago HP camera with a high resolution collimator. This combination is ideally suited to the physical properties of ^{99m}Tc , since the lymph nodes are much like point sources. As a result, satisfactory studies may be obtained even with lower count rates simply by adjusting intensity. Multiple views are simple to obtain using the cathode-ray tube display to determine optimum patient positioning. The pinhole collimator may be used to separate apparently confluent node groups and is another advantage of the camera. Results of tomography studies using the Nuclear-Chicago system have not been encouraging to date (Figure 5). The manner in which the images are generated places a negative area

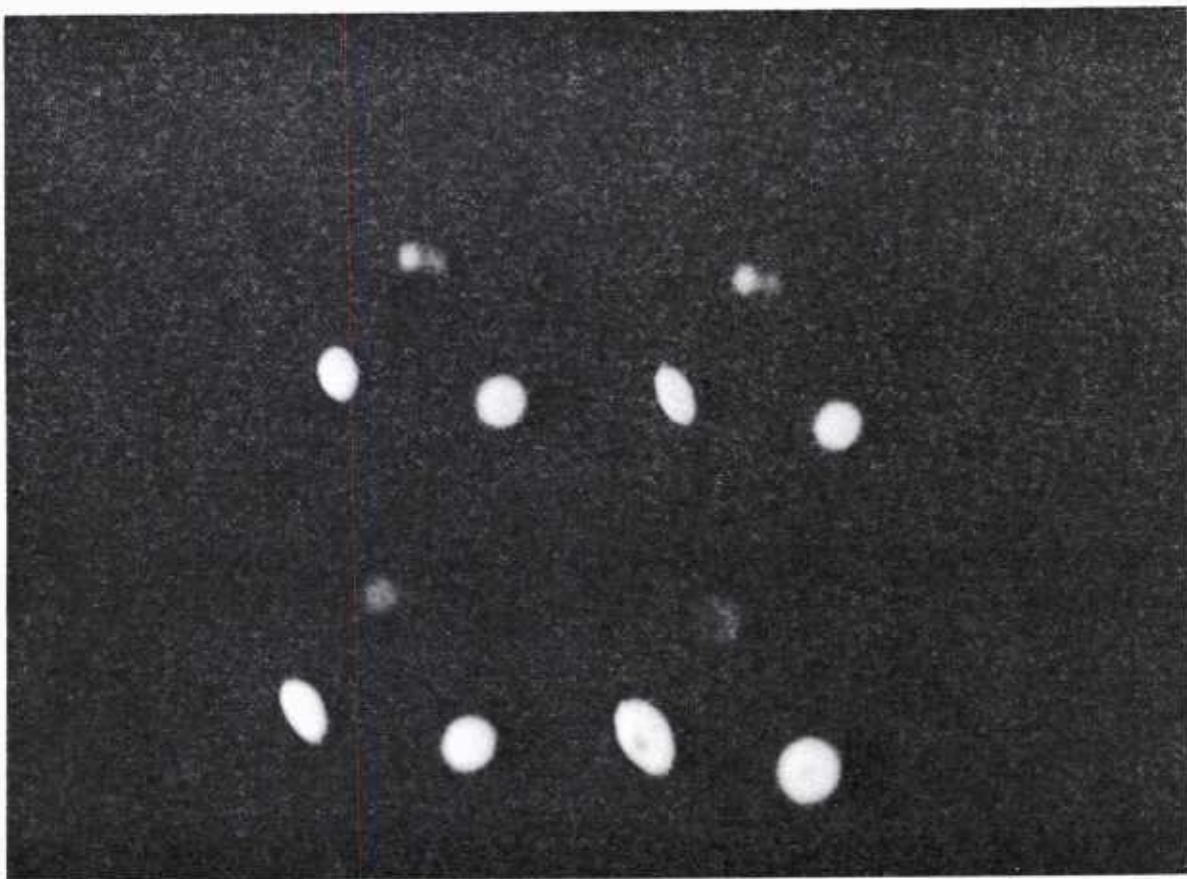


Figure 5. Respective tomographic cuts of the ilioinguinal lymph node chain in dogs

in the middle of a point source when it is out of the focal plane. Although in isolated cases tomography may prove useful, the point source nature of the lymph nodes makes this potentially misleading.

As indicated by the time activity curves in Figure 3, the dose to scan time is flexible; however, there is some advantage in scanning before recirculation occurs, e.g., at 90 to 150 minutes after injection. Potentially confusing background activity in liver, bowel and genitourinary organs is thereby avoided, but this has not been a major problem. Table II is a summary of our techniques for pedal studies.

Table II. Technique for Pedal Study

Dosage	Injection site	Additional medications	Exercise	Dose to scan time	Views obtained
2.0 mCi per foot	Subcutaneous, medial web spaces	None	Ambulation, if able; passive exercise otherwise	90 - 150 minutes	<ol style="list-style-type: none">1. Anterior pelvis (ilioinguinal nodes)2. Anterior abdomen (periaortic nodes)3. Obtain additional views (i.e., supraclavicular, tomographic) as indicated

Dosimetry. Dosimetric calculations are shown in Table III. A dose of 2 mCi per site has proven adequate for imaging although higher activities may be safely given. Biological assumptions were that 20 percent of the administered dose would remain at the injection site and the rest would migrate through the lymphatics. Colloid reaching the systemic venous return localizes in the reticuloendothelial system. The absorbed dose to individual lymph nodes will vary depending on the percent localization and the size of the node. There are approximately 350 g of abdominopelvic lymphatics.

Table III. Radiation Dose Calculations

2 mCi ^{99m}Tc minicolloid injected subcutaneously in each foot; 20 percent retained at injection site					
Expected migration of the radiopharmaceutical: feet \rightarrow lymph nodes \rightarrow blood \rightarrow RES					
Organ	Percent uptake	μCi	Mass (g)	\bar{g}	Maximum dose (rads)
Whole body	100	4000	70,000	126	<0.1
Injection site	20	400	5	10	29.0
Gonads	1	40	40	10	0.4
Liver	100	4000	1,700	61	1.5
Lymph					
Case 1*	1.4	57	5	10	4.1
Case 2†	10	400	5	10	29.0

* Single node, uniform distribution

† Single node, 10 percent total dose

IV. CONCLUSION

Most of the radionuclide lymphography experience in the past has been with gold-198 colloid.^{4, 6, 9, 11, 13} Despite a large European series and optimistic American reports, the clinical use of this radiopharmaceutical has not found general acceptance.^{1, 4, 9}

The high beta radiation dose from gold-198 is a distinct disadvantage which precludes the use of more than 100 μCi on each side for pedal studies. The rather high principal gamma energy (411 keV) may result in inefficient collimation with additional loss in spatial resolution and sensitivity. Previous studies have shown that the amount and distribution of activity from this radiopharmaceutical appear to be inadequate for resolution of individual nodes especially in the periaortic areas.^{6, 9} The required 24-hour dose to scan time is another disadvantage of gold-198 radiocolloid. Since imaging cannot be performed on the same day, this makes the procedure more cumbersome and precludes single visit outpatient studies.

The ideal physical properties of ^{99m}Tc labeled products and the smaller patient dose allow the use of greater amounts of the radiopharmaceutical and achievement of better resolution. Individual lymph nodes may be seen as high as the periaortic region. Dose to scan time is short, which permits single visit studies.

The greatest common advantages of radionuclide lymphographic techniques derive from their simplicity. Only 5 minutes of the physician's time is required to inject the radiopharmaceutical through the subcutaneous or submucosal route. The remaining technician and equipment time needed to complete the procedure varies but is uniformly less than that required for studies involving radiographic contrast media. Complications are virtually nonexistent.^{3,5,6} The radionuclide techniques may be used in anatomical areas where adequate lymphatic vessels for cannulation are not present. These procedures also represent a more physiologic approach for studying lymphatic dynamics.

Most importantly, serial repetitions may be as easily carried out as the initial study. This is a distinct advantage over radiographic lymphograms in the long-term follow-up of patients. The relatively poor resolution is partially offset by the ability to assess change from one study to the next. The results of this study support the observations of others that assessment of the accuracy of radionuclide scanning must come from direct clinical observation and nodal biopsy rather than from comparison with radiographic lymphography. Radiographs have their own peculiar inaccuracies and are subject to significant variations in interpretation.^{4,6}

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13. ABSTRACT

The technique for lymphatic system evaluation by imaging with technetium-99m "minicolloid" is presented. Results from studies which compared the smaller particle radiocolloid with the commercially available radiopharmaceutical of the larger colloid particle size demonstrated the former to be a far superior agent for imaging the lymphatic system. This appears to be due to its more favorable migration through successive nodal groups. Other proven advantages of the minicolloid material were same-day imaging, lower absorbed dose, and better resolution. Radionuclide lymphography is expected to play an increasing role, in conjunction with radiographic lymphography, as larger numbers of patients suspected of lymphatic system disease are evaluated using the nuclear medicine techniques and approaches described.